

APPARATUS FOR AUTOMATED PROCESSING BIOLOGICAL SAMPLES

TECHNICAL FIELD

5 The present invention relates to a method and an apparatus for automatic processing of at least one biological sample accommodated on a carrier member, such as microscopic slide, by applying a predetermined amount of reagents in a sequence according to a processing protocol, wherein at least one slide is provided in a slide rack assembly.

10 This application relates to the field of sample processing systems and methods of processing samples. The present invention may be directed to the automated processing, treatment, or even staining of samples arranged on carriers, such as microscope slides, and in some embodiments, directed to the continuous or batch processing of samples and
15 carriers, as well as washing elements of a sampling system. Embodiments may further relate to control systems for sample processing and data acquisition, data maintenance, and data retrieval for sample processing. Applications to which the present invention may especially relate include cytology, immunohistochemistry, in-situ hybridization, fluorescent in-situ hybridization, special staining, and microarrays, as well as potentially
20 other chemical and biological applications.

BACKGROUND

25 The biological sample may suitably be selected from histological material, including formalin fixed and paraffin embedded material, cytological material, fine needle aspirates, cell smears, exfoliative cytological specimens, touch preparations, bone marrow specimens, sputum samples, expectorates, oral swabs, laryngeal swabs, vaginal swabs, bronchial aspirates, bronchial lavage, gastric lavage, and body fluids. Such may be subjected to various treatments.

30 Sample processing in immunohistochemical (IHC) applications and in other chemical and biological analyses may require one or a number of various processing sequences or protocols as part of an analysis of one or more samples. The sample processing sequences or protocols may be defined by the individual or organization requesting an analysis, such

as a pathologist or histologist of a hospital, and may be further defined by the dictates of a particular analysis to be performed.

5 In preparation for sample analysis, a biological sample may be acquired by known sample acquisition techniques and may comprise, for example in IHC applications, tissues generally or even in some applications one or a plurality of isolated cells, such as in microarray samples, and may be presented on a sample carrier such as a microscope slide. Furthermore, the sample may be presented on the carrier variously and potentially in some form of preservation. As one example, a human biopsy sample may be fixed and
10 embedded in a suitable media like paraffin or epon, before being mounted onto a carrier. The sample may be treated according to the protocol, which may include the following non limiting procedural steps: deparaffination, antigen retrieval, denaturing, washing, incubation with various immunological reagents, molecular probes or dyes, stringency wash or counterstaining.

15 Histological, cytological, ISH, IHC special stains and other applications, for example, may require processing sequences or protocols that comprise many and laborious steps such as deparaffinization, target retrieval, and staining. Such steps are not special for ISH procedures, but goes for IHC, cytological, specials or ISH stainings. Previously, in some
20 applications, these steps may have been performed manually, potentially creating a time-intensive protocol and necessitating personnel to be actively involved in the sample processing. Attempts have been made to automate sample processing to address the need for expedient sample processing and a less manually burdensome operation. However, such previous efforts may have not fully addressed the needs for an automated sample
25 processing system. Previous efforts to automate sample processing may be deficient in several aspects that prevent more robust automated sample processing, such as: the lack of sufficient computer control and monitoring of sample processing; the lack of information sharing for processing protocol and processing status, especially for individual samples; the lack of diagnostic capabilities; and the lack of real-time or adaptive capabilities for
30 multiple sample batch processing.

From US Patent No. 5,839,091 an automated sample processing for samples presented on carriers such as slides is known. In this apparatus, an array of slides is stained by a robotic device delivering reagents onto the slides. The staining apparatus is provided with a lid
35 cover, which provides a protective cover of the biological samples on the carriers in the

apparatus during the staining processes as well as containing stains of the some times hazardous reagent materials within the apparatus and preventing operators from being exposed to such stains.

- 5 Slides can be any suitable solid or semi solid support for the biological sample. In particular, the support may be a microscope slide, a membrane, a filter, a polymer slide, a chamber slide, a dish, or a petridish.

Some staining processes involve the use of hazardous materials, such as toxic materials.

- 10 These materials may be collected in special containers in order to ensure safe handling of the waste material. However, this does not sufficiently protect the laboratory environment in which the apparatus is placed from being contaminated with toxic material. Moreover, in some staining processes or other treatments in the apparatus heat is applied. This increases the risk of vaporising reagents which then may escape to the outside of the
15 apparatus.

- In the apparatuses known in the art, a protective hood or similar plastic cover is put over the staining apparatus in order to shield off the biological samples during the staining. In this known technique, one risk is the drying out of slides and lack of control of airspeed
20 and temperature.

DISCLOSURE OF INVENTION

- On this background, it is an object of the invention to provide an apparatus for automatic
25 staining of biological samples, in which the hazardous materials and toxic or otherwise damaging fumes are contained and automatically handled, so that operators need not be involved in the handling of such types of materials.

- The invention consists of a method and an apparatus for automatic staining at least one
30 biological sample accommodated on a carrier slide by applying a predetermined amount of reagents in a predetermined sequence according to a staining protocol, said apparatus comprising: a housing frame; at least one staining section for accommodating at least one slide, said at least one staining section is provided within said housing; a hood cover protecting said at least one staining section in said housing; wherein the hood cover
35 completely encloses the staining section defining an interior space; and wherein the

apparatus further comprises climate control device that provide control of the environment within said interior space; and sensor device to provide feedback signals to the climate control means.

5 In the staining apparatus, fumes from the reagents and other processing liquids used in the apparatus for performing the treatment of the biological samples are present. In an apparatus according to the invention, these fumes may be exhausted from the interior space of the apparatus, just as the climate in the apparatus may be controlled. In particular, in order to avoid the volatile fumes from escaping into the surroundings, the climate
10 control device preferably includes a pressure control device that can ensure a slight sub-pressure within the interior space. By maintaining a slight sub-pressure inside the apparatus, the fumes are kept inside in the interior space, from where the climate control system may remove the volatile fumes. These fumes may advantageously be collected in suitable storage or otherwise be disposed of.

15 Advantageously, the climate control device may also include humidity control within the interior space. This makes an apparatus according to the invention more suitable for some special sample treatment applications, just as the climate requirement inside the laboratory where the apparatus is placed may be less strict. Moreover, the climate control device may
20 preferably also include a temperature control device that can control the ambient temperature of the air within the interior space.

Climate control, especially control of temperature and humidity is important for ensuring standardized and reproducible staining protocols, as the biological samples can easily dry
25 out or changes properties, including morphology and antigenicity. Also, the controlled environment can ensure uniform chemical reaction conditions on the slides during the staining protocol.

The climate control device in an apparatus according to the invention may include an
30 exhaustion device that can remove fumes from the interior space. These exhaustion devices are preferably adapted to draw air from an outlet positioned below the level in which the at least one slide is accommodated. Hereby, fumes are drawn away from the hood and the slides. This may reduce any risk of cross-contamination. Cross contamination could be a problem, as some fumes can adsorb to the biological sample,
35 resulting in a change of properties. Especially hydrophobic, acidic, basic, strong

chaotropic or otherwise reactive or corrosive fumes can cause serious and unwanted cross contamination.

5 The sensor device is preferably adapted to sense at least one climate parameter from the group comprising temperature, pressure, humidity, airspeed and the presence of toxic elements in fume. These measured air characteristics are used for manipulating the inflowing air into the interior space, so that it is ensured that the biological samples are processed in a controlled ambient environment and that it is ensured that the samples do not dry out or are otherwise being deteriorated.

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The sensor device may comprise internal sensors located inside the interior space. Alternatively, the sensor may comprise external sensors located outside the interior space, such as at or inside an air inlet/outlet manifold, in a laboratory facility accommodating the apparatus, or outside the building accommodating the laboratory.

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In the preferred embodiment, the cover is at least one openable hood. The cover is an integrated part of the apparatus, e.g. a hinged hood which may be swung open. An automatic lock may be provided to prevent any opening during the processing.

20 In an embodiment, the cover is a plurality of covers arranged to cover a plurality of sections of the apparatus, such as at least one biological sample accommodated on a carrier in the at least one processing section. Hereby, different environments may be created for different samples. A plurality of interior spaces may be defined by a plurality of covers in the apparatus, each interior space including at least one section arranged for
25 comprising at least one sample on a carrier and/or at least one section arranged for comprising at least one reagent in a container.

The climate control device may be arranged to controlling the climate in each interior space, comprising at least one sample on a carrier, according to a sample processing
30 protocol defined for that particular sample. In an embodiment of the invention, the climate control device receives input signals from internal and/or external sensors, and are arranged to control the climate in each interior space, according to the input signals. The climate control device may then preferably connected for data communication with a data processing device, such as a computer, wherein the protocol for the processing of the
35 particular sample is stored, and where from control data are provided to the climate control

device. Hereby, an automatic climate control is provided for the sample processing, whereby it may be possible to provide a selected air environment for a particular sample processing protocol.

- 5 In a preferred embodiment of the invention, the hood cover is provided with one or more seal elements to provide an air-tight seal between the cover and the housing. Hereby, the sub-pressure need not be present or a higher pressure inside may be provided for achieving a more precise climate control inside the apparatus.
- 10 The exchange of air between the interior space and the surroundings may be carried out via an inlet and an outlet. In an embodiment of the invention, the inlet may be provided for supplying air into the interior space including an air inlet opening in the housing, and wherein air manipulation device are provided in addition to said inlet means to adapt the inflowing air with predetermined characteristics. Hereby, the inside climate of the air in
- 15 the interior space may be accurately controlled.

According to the present invention, a method and an apparatus is provided which allows air containing the fumes to be removed to the exterior or removed on a filter device before the cleaner air is being recycled.

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Also, it should be understood, that the air drawn into the interior space in addition to be temperature controlled by heating or cooling, also may be added humidity by spraying water droplets or using a filter device, or added other components, like nitrogen gas, carbondioxide or inert gasses to control the environment in the hood.

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In one preferred embodiment, the inlet air is drawn through a humid filter device to ensure high and uniform humidity in the chamber.

In another preferred embodiment, the humidity is controlled by spraying water droplets or

30 having a water surface.

In yet another preferred embodiment, recycled air is drawn through filters to remove fumes and filters to adjust the humidity.

In yet another preferred embodiment, the humidity is controlled to never be below a predetermined level, to prevent drying out of the sample.

Also, disinfectants, UV protectants or other compounds could be added to the inlet air to prevent microbial growth or discolouring.

It should be understood that by controlling the environment by the present invention, the mechanical and electrical parts are also protected from corrosion and wear. Also, by an apparatus according to the invention the air may be moved around inside the interior space which ensures uniform and controllable temperature control.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention is described with reference to a preferred embodiment with reference to the drawings, in which:

- Fig. 1 is a schematic perspective view of a staining apparatus according to the preferred embodiment of the invention;
- Fig. 2 is a top view of the work area in the staining apparatus shown in fig. 1;
- Fig. 3 is a schematic side view of the staining apparatus of fig. 1; and
- Fig. 4 is an example of a ventilation assembly mountable in the housing in an apparatus according to the invention.

BEST MODES FOR CARRYING OUT THE INVENTION

A staining apparatus 1 according to the invention is shown in the figures. The staining apparatus 1 comprises a rectangular housing frame 4 surrounding a reagent station 2 comprising an array of reagent bottle compartments wherein each compartment a reagent vial 3 is placed, and a first and second slide sections 5 wherein a number of separate rack assemblies 11 is placed, potentially in some embodiments in one or more drawer assemblies 6, and where each rack assembly 11 accommodates a number of microscope slides 7 mounted side by side in the rack assembly. In the embodiment shown, each rack may hold up to 8 slides, but the rack may be designed to hold any suitable number of slides. With eight racks arranged side by side, the shown embodiments may hold up to 64

slides 7 each having a biological sample, e.g. a tissue mounted on the upper side of the slide, so that reagent may be applied from above to the sample on each slide.

5 A robot arm 20 for moving a probe 10 in X and Y (as well as Z) direction as indicated by the arrows X and Y is arranged above the frame 4 of the staining apparatus. The robot arm 20 may therefore position the probe 10 above all reagent vials 3 as well as above all the microscope slides 7, and may further operate the probe 10 to aspirate portions of reagent contained in any of the vials 3, to transfer the portion of reagent and apply it to any of the slides 7 in order to provide a selected staining or treatment of the sample on each slide 7.
10 By use of suitable control, e.g. a computer (not shown) having the appropriate software and input data for the purpose, this staining apparatus 1 is able to automatically stain or treat samples requiring different staining or treatment reagents and processes.

As shown in fig. 1, the probe 10 is accommodated in a robotic head 22 and is manipulated
15 by the robot arm 20. The probe 10 is raised to an upper position (in a Z direction) where it is clear of the vials 3 underneath the probe 10, but the robot comprises means in the robotic head 22 for lowering the probe 10 in order to dip the probe tip into the content of a selected reagent vial 3 and to aspirate a selected amount of reagent for the selected staining or treatment process. The robotic head 22 may also be provided with a CCD
20 camera 25, in some embodiments configured to point downwards. The camera is utilised to determine status information of the slides and the reagent bottles and other features of the apparatus in the work area, for example reading information which may or may not be encrypted, that is provided on a reagent container to determine the reagent type and the reagent location within the system. The camera may also determine status of the tissue
25 sample carriers, for example the location of a particular slide, informational indicia, such as which may be converted into an encrypted format, that indicate information about the tissue sample presented on the slide or the processing protocol to be performed.

The staining apparatus 1 of the present embodiment further comprises a probe washing
30 station 8 and a reagent mixer 9, and the robot arm 20 is furthermore arranged to transfer the probe 10 to the washing station 8 as well as to the reagent mixer 9.

As shown in fig. 3, the apparatus 1 is provided with an openable hood cover 40, which is pivotably attached to the frame housing 4. The hood 40 is shown in a closed position and
35 an open position 40' is indicated in dotted lines in the figure.

In the bottom of the frame housing 4, the slide rack assemblies 11, potentially in some embodiments in one or more drawer assemblies 6 are provided. An interior space 120 is defined by this hood cover 40 and the frame 4. The staining sections 5 and the reagent station 2 are arranged such that they are accessible by the robotic arm 20 via the equipment arranged on the robotic head 22.

A climate control device 121 is provided for controlling the pressure and potentially also the temperature and the humidity of the air in the interior space 120 inside the apparatus 1. This climate control 121 includes an outlet 122 and an inlet 123 allowing for an exchange of air in a controlled manner. Exhausted air from the interior space may be directed to a collection storage or disposed of in other manners depending on the requirements on the location of the apparatus. The exhausted air may – in particular in some staining or other processes – include volatile fumes or other toxic or unwanted fumes from the reagents and other liquids used for the processing of the biological samples.

The ambient air in the interior space 120 is drawn through the rack assembly 11 of the drawer assembly 6 in the staining sections 5. In the drawer assembly 6, other processing devices are preferably also provided, such as pretreatment tanks in which the biological specimens arranged on slides may be immersed in a pretreatment liquid which is often heated to a processing temperature. By drawing the fumes out of the interior space 120 at a location close to the heating sources and below the level in which the slides are arranged, the fumes are essentially prevented from diffuse to widely in the interior space, whereby the risk of such fumes coming into unwanted contact with biological samples, other reagents or processing liquids may be avoided.

Besides maintaining a slight sub-pressure inside the hood, the climate control may also be adapted to controlling the temperature and the humidity inside the apparatus, so that such climatic conditions do not interfere with the staining processes. Hereby, the apparatus may easily be adapted to changes in the environment, such as seasonal changes, temperature changes, just as the apparatus according to the invention may be able to adapt itself to a specific location, e.g. adapt to difference in altitude, etc.

The temperature, humidity, airflow rate and/or other environmental factors can be controlled by a feed back mechanism from a sensor device, such as one or more sensors arranged in the hood or elsewhere inside the interior space and/or external sensors, which

may be advantageous in order to compensate for external influences such as high temperature or extremely dry climate in local areas having extreme climate variations.

- 5 The system may further have the ability to vent toxic and or flammable fumes from inside the cabinet portions or enclosure to exit ports that can be connected to an external system vent or hood, such as vent 160. Embodiments may comprise exit ports from the internal enclosure of the system and exit ports from the bulk fluid containers of the system, such as reagent containers. Fumes may be isolated and removed from various compartments
10 within the system. An environmental control system addresses the system's ventilation to ensure the evacuation of volatile organic vapours, keeping the concentration of these materials below established toxic and explosive limits.

In some embodiments, a ventilation system such as that shown in Figures 3 and 4 will
15 draw hazardous fumes from the vicinity of each processing tank in the drawer assemblies 6 by an exhaustion device that may comprise dedicated ducts 122, 123. These ducts will feed a manifold 121 running along the rear of the instrument. A separate ventilation system for an electronic chassis of the system may be similarly isolated having independent air input and output vents. Each of these systems has independent air input
20 and output air vents. At one end of the manifold, a centrifugal blower will exhaust the fumes to an exit port that may be connected to the facility hood/exhaust system.

In fig. 4, a ventilation assembly 80 for mounting in the apparatus is shown. Through this ventilation assembly 80 air may be exchanged between the interior space 120 and the
25 surroundings. In addition to the inlet and outlet ducts 122, 123, air manipulation devices 81, 82 may be arranged. These air manipulation devices may include moisture control, air pressure and air flow regulation in order to control the environment in the interior space automatically in response to sensor readings inside the interior space.

- 30 Above, the apparatus according to the invention is described according to some preferred explanatory embodiments. However, it is realised by the invention that many other variations and equivalents of the apparatus may be carried out without departing from the scope of the invention as specified in the accompanying claims.

As can be easily understood from the foregoing, the basic concepts of the present invention may be embodied in a variety of ways. It involves both sample processing techniques as well as various systems, assemblies, and devices to accomplish sample processing, input, and other functions. In this application, the sample processing techniques are also disclosed as part of the results shown to be achieved by the various systems, assemblies, and devices described and as steps which are inherent to utilization. They should be understood to be the natural result of utilizing the devices as intended and described. In addition, while some devices are disclosed, it should be understood that these not only accomplish certain methods but also can be varied in a number of ways. Importantly, as to all of the foregoing, all of these facets should be understood to be encompassed by this disclosure.

The discussion included in this application is intended to serve as a basic description. The reader should be aware that the specific discussion may not explicitly describe all embodiments possible; many alternatives are implicit. It also may not fully explain the generic nature of the invention and may not explicitly show how each feature or element can actually be representative of a broader function or of a great variety of alternative or equivalent elements. Again, these are implicitly included in this disclosure. Where the invention is described in device-oriented terminology, each element of the device implicitly performs a function. Importantly, neither the description nor the terminology is intended to limit the scope of the claims which may be included at any time.

It should also be understood that a variety of changes may be made without departing from the essence of the invention. Such changes are also implicitly included in the description. They still fall within the scope of this invention. A broad disclosure encompassing both the explicit embodiment(s) shown, the great variety of implicit alternative embodiments, and the broad methods or processes and the like are encompassed by this disclosure and may be relied upon at any time.

Further, each of the various elements of the invention and claims may also be achieved in a variety of manners. This disclosure should be understood to encompass each such variation, be it a variation of an embodiment of any apparatus embodiment, a method or process embodiment, or even merely a variation of any element of these. Particularly, it should be understood that as the disclosure relates to elements of the invention, the words for each element may be expressed by equivalent apparatus terms or method terms -- even

if only the function or result is the same. Such equivalent, broader, or even more generic terms should be considered to be encompassed in the description of each element or action. Such terms can be substituted where desired to make explicit the implicitly broad coverage to which this invention is entitled. As but one example, it should be understood that all actions may be expressed as a means for taking that action or as an element which causes that action. Similarly, each physical element disclosed should be understood to encompass a disclosure of the action which that physical element facilitates. Regarding this last aspect, as but one example, the disclosure of a "retention element" should be understood to encompass disclosure of the act of "retaining" -- whether explicitly discussed or not -- and, conversely, were there effectively disclosure of the act of "retaining", such a disclosure should be understood to encompass disclosure of a "retention element" and even a "means for retaining". It should also be understood that in jurisdictions where specific language may be construed as limiting, as but one example in the United States where some interpretations of "means for" elements can be construed narrowly, broader equivalent language may be used and should be understood as encompassed by this specification. Such changes and alternative terms are to be understood to be explicitly included in the description.

Any patents, patent applications, publications, or other references mentioned in this application for patent are hereby incorporated by reference. In addition, as to each term used it should be understood that unless its utilization in this application is inconsistent with such interpretation, common dictionary definitions should be understood as incorporated for each term and all definitions, alternative terms, and synonyms such as contained in the Random House Webster's Unabridged Dictionary, second edition are hereby incorporated by reference as well as the definitions presented by searchStorage.com, such to be considered as representing the meaning of the terms as understood by computer professionals. Finally, any priority case for this application is hereby appended and hereby incorporated by reference.

Thus, the applicant(s) should be understood to have support to claim at least: i) each of the sample processing systems and subsystems as herein disclosed and described, ii) the related methods disclosed and described, iii) similar, equivalent, and even implicit variations of each of these systems, assemblies, devices and methods, iv) those alternative designs which accomplish each of the functions shown as are disclosed and described, v) those alternative designs and methods which accomplish each of the functions shown as

are implicit to accomplish that which is disclosed and described, vi) each feature, component, and step shown as separate and independent inventions, vii) the applications enhanced by the various systems or components disclosed, viii) the resulting products produced by such systems or components, and ix) methods and systems, assemblies, devices, and apparatuses substantially as described hereinbefore and with reference to any of the accompanying examples, x) the various combinations and permutations of each of the elements disclosed, xi) each potentially dependent claim or concept as a dependency on each and every one of the independent claims or concepts presented, xii) processes performed with the aid of or on a computer as described throughout the above discussion, xiii) a programmable system as described throughout the above discussion, xiv) a computer readable memory encoded with data to direct a computer comprising means or elements which function as described throughout the above discussion, xv) a computer configured as herein disclosed and described, xvi) individual or combined subroutines and programs as herein disclosed and described, xvii) the related methods disclosed and described, xviii) similar, equivalent, and even implicit variations of each of these systems and methods, xix) those alternative designs which accomplish each of the functions shown as are disclosed and described, xx) those alternative designs and methods which accomplish each of the functions shown as are implicit to accomplish that which is disclosed and described, xxi) each feature, component, and step shown as separate and independent inventions, and xxii) the various combinations and permutations of each of the above.

Further, if or when used, the use of the transitional phrase "comprising" or the like is used to maintain the "open-end" claims herein, according to traditional claim interpretation. Thus, unless the context requires otherwise, it should be understood that the term "comprise" or variations such as "comprises" or "comprising" or the like, are intended to imply the inclusion of a stated element or step or group of elements or steps but not the exclusion of any other element or step or group of elements or steps. Such terms should be interpreted in their most expansive form so as to afford the applicant the broadest coverage legally permissible.

Any claims set forth at any time are hereby incorporated by reference as part of this description of the invention, and the applicant expressly reserves the right to use all of or a portion of such incorporated content of such claims as additional description to support any of or all of the claims or any element or component thereof, and the applicant further

expressly reserves the right to move any portion of or all of the incorporated content of such claims or any element or component thereof from the description into the claims or vice-versa as necessary to define the matter for which protection is sought by this application or by any subsequent continuation, division, or continuation-in-part application thereof, or to obtain any benefit of, reduction in fees pursuant to, or to comply with the patent laws, rules, or regulations of any country or treaty, and such content incorporated by reference shall survive during the entire pendency of this application including any subsequent continuation, division, or continuation-in-part application thereof or any reissue or extension thereon.

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